MRID NO. 448073-29

# DATA EVALUATION RECORD FISH LIFE-CYCLE TOXICITY TEST **GUIDELINE 72-5**

**CHEMICAL:** Fluazinam PC Code No.: 129098

Purity: 97.4% Technical Fluazinam TEST MATERIAL:

(IKF-1216)

3. CITATION:

> Author: Emily Dionne

Technical Fluazinam (IKF-1216) - The <u>Title:</u>

Chronic Toxicity to the Fathead Minnow (Pimephales promelas) During a Full

Life-Cycle Exposure

Study Completion Date: July 25, 1995

Springborn Laboratories, Inc., Wareham, Laboratory:

<u>Laboratory Report ID</u>: 94-1-5123

Sponsor: Ishihara Sangyo Kaisha, Ltd., Tokyo,

MRID No.: 448073-29 DP Barcode: Not reported

Karl Bullock, M.S., Environmental Scientist, REVIEWED BY:

Golder Associates Inc.

Kal bullet

Date: 10/13/99 Signature:

APPROVED BY: Pim Kosalwat, Ph.D., Senior Scientist,

Golder Associates Inc.

signature: P. Kosalwat Date: 10/13/99

APPROVED BY: III Rextrole Ph.D. Senior Aquatic Diologist 1

Date: 1/10/2001 Signature:

**Study Parameters:** 

Test Species: Pimephales promelas Age of Test Organism: <24 hours old embryos

Test Duration: 278 days Study Method: Flow-Through
Type of Concentrations: Mean measured

**CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirements for a fish full lifecycle toxicity test using fathead minnows. Based on mean measured concentrations, the LOEC and NOEC for fathead minnows exposed to Technical Fluazinam were 1.4 and 0.69 ppb, respectively. The geometric mean MATC was estimated to be 0.98 ppb.

# Results Synopsis:

Most Sensitive Endpoint: F, hatching success

NOEC: 0.69 ppb LOEC: 1.4 ppb MATC: 0.98 ppb

### 8. ADEQUACY OF THE STUDY:

A. Classification: Core.

B. Rationale: Fulfills guideline requirements.

C. Repairability: N/A

# 9. GUIDELINE DEVIATIONS:

- 1. Dilution water hardness (25-40 mg/L as  $CaCO_3$ ) and pH (6.9-7.2) were slightly less than recommended (40-48 mg/L as  $CaCO_3$ , pH range of 7.2-7.6).
- 2. The  $F_1$  larval exposure period (30 days) was less than recommended (8 weeks).
- 3. Illumination (377 to 1292 lumens) was greater than recommended (10 100 lumens).
- 4. Larval chamber construction material and drain height were not reported.
- 5. The volume of the embryo incubation cups was not reported.
- 6. During test days 161 278, test aquaria were aerated.
- 7. Only two replicates were utilized; guidelines recommend using four replicates.

# 10. SUBMISSION PURPOSE:

# 11. MATERIALS AND METHODS:

# A. Biological System:

Guideline Criteria	Reported Information
Species: Prefer sheepshead minnow (Cyprinodon variegatus) or fathead minnow (Pimephales promelas).	Pimephales promelas
Source and acclimation	Embryos were obtained from in- house brood stock. Fish were held under conditions similar to testing.
Age at beginning of test: Embryos 2 to 24 hours old	<24 hours old
Feeding: Fish should be fed at least twice daily and should not be fed for at least 24 hours prior to test termination.	Newly hatched fry were fed live brine shrimp nauplii three times daily for the first 26 days. Between days 27 and 29, small amounts of Zeigler Brothers Prime flakes were also offered once daily. Beginning on day 30 (post-hatch), fish were fed twice daily during weekdays, once with frozen brine shrimp and once with flake food. On weekends, fish were fed once daily. Feeding was withheld for 24 hours prior to test termination.

Guideline Criteria	Reported Information
Embryo Exposure (Four-Five Days): Embryos (≤24 hours old) from at least 3 separate spawns should be randomly distributed to embryo cups.	Embryos (<24 hours old) from 18 spawns tiles were randomly distributed to embryo cups.
A minimum of 50 embryos (≤24 hrs old) per replicate cup, 4 cups per treatment should be used.  Parameters measured: Survival of embryos Time required to hatch Hatching success Survival of fry for 4 weeks	50 embryos per cup (randomly assigned five at a time); 2 cups per replicate test chamber; 2 replicate chambers per treatment and control.  All parameters listed at left were measured.
Dead and fungused embryos should be counted and removed daily.	Dead embryos were recorded and removed daily until hatching was complete.
Larval-Juvenile Exposure (From Hatch to 8 Weeks): After hatching, each group of larvae is randomly reduced to a minimum of 25 fish and released in replicate larval growth chambers. The random selection must include any fish that are lethargic or deformed.	95% hatch was complete on Day 5. Larvae from the incubation cups in each control and treatment chamber were impartially reduced to 50 per replicate on Day 5 and released into the larval growth chambers on Day 6 (25 fish per growth chamber, 2 growth chambers per replicate). At 66 days post-hatch, juveniles were impartially reduced to 25 per
Parameters measured: Fish survival (determined by counting the number of live fish in each replicate growth chamber weekly).	replicate.  Both parameters at left were measured.
<ul> <li>Total lengths (mm) of all fish at 4 and 8 weeks after hatching.</li> </ul>	

#### Guideline Criteria Reported Information Juvenile-Adult Exposure Spawning tanks (2 per treatment) (From 8 wks posthatch to were divided into 3 equal the end of the spawning spawning compartments using phase [32-40 wks]): nylon mesh screen dividers. On Day 161, one male and two At 20-24 weeks after females were distributed to each hatching, mature fish are spawning compartment. The placed in a spawning tank remaining adults were left in of the same concentration the exposure aquaria until test (4 males and 4 females termination (Day 278). Spawning randomly chosen and substrate consisted of 10-cm assigned). The spawning sections of 4"-diameter PVC tank is divided into 4 pipe, which were halved and individual spawning placed concave surface down to form "arches". Two such chambers with appropriate spawning substrates. spawning substrates were provided to each spawning group. The substrates are examined Substrates were examined, and daily and embryos removed, embryos were removed and counted counted, and recorded daily. separately for each pair. For fathead minnow, adult The spawning phase was exposure should be terminated at 278 days postterminated when no spawning hatch. occurs for one week. For sheepshead minnow, testing should be terminated after spawning is observed for 2 weeks.

# Second Generation Embryo Exposure (4-5 days):

50 embryos from each conc. level are randomly selected and transferred to incubation cups for hatch. Use the same test procedures as those for parental generation.

Embryos not selected are discarded.

10-15 sets of 50 embryos from each replicate (average of 740 embryos per replicate at test concentrations which did not affect reproduction) were incubated and the percent hatch was determined.

Embryos not selected were kept for residue analysis.

Guideline Criteria	Reported Information			
Second Generation Larval- Juvenile Exposure (From Hatch to 4-8 wks): After hatching, 25 larvae are released in each growth chambers (2 chambers per treatment).	An average of 6 groups (3 per replicate) of 25 larvae from test concentrations which did not affect reproduction were released into growth chambers for each treatment.			
Each group of 2 <sup>nd</sup> generation fish is terminated 8 wks after hatching.	After 30 days of post-hatch exposure, the 2 <sup>nd</sup> generation fish were terminated.			
Fish are blotted, weighed, and measured before being discarded.	Fish were measured for individual length and wet weight.			

Comments: None.

# B. Physical System:

Guideline Criteria	Reported Information
Test Water:  Sheepshead Minnow  1. Natural seawater   (sterilized and filtered)   or a commercial mixture.  2. Natural seawater with a   salinity of ≥15 parts per   thousand (weekly range of   salinity <6% and monthly   pH range <0.8 pH units).  Fathead Minnow  1. Reconstituted water or   water from unpolluted   well or spring   (sterilized and tested   for pollutants).  2. Hardness of 40 to 48 mg/L   as CaCO <sub>3</sub> and pH of 7.2 to   7.6.	<ol> <li>Well water, aerated and passed through an ultraviolet sterilizer before delivery to the proportional diluter. Water was screened semiannually for contaminants and monthly for TOC</li> <li>The hardness ranged from 24 to 30 mg/L as CaCO<sub>3</sub>. The pH ranged from 6.7 to 7.6.</li> </ol>

Guideline Criteria	Reported Information
Test Temperature: Fathead: 25°C and should not remain outside the range of 24 to 26°C for more than 48 hours.  Sheepshead: 30°C.	Range: 22 - 26°C  The temperature of 23°C did not occur for any period > 48 hours.  The low temperature of 22°C occurred once for a period of <24 hours.
Photoperiod: 16-hour light/8-hour dark. Light intensity of 10-100 lumens at water surface.	Graduated photoperiod based on dust to dawn times in Evansville, IN. Range of 10.3 to 15.45 hours of light/day.  377 to 644 lumens/m² for exposure aquaria, 969 to 1292 lumens/m² for spawning aquaria.
<ul> <li>Dosing Apparatus:</li> <li>1. Intermittent flow proportional diluters or continuous flow serial diluters.</li> <li>2. A minimum of 5 toxicant concentrations with a dilution factor ≤0.5.</li> <li>3. One control should be used.</li> </ul>	<ol> <li>Intermittent flow proportional diluter.</li> <li>Five with a dilution factor of 0.5.</li> <li>A dilution water control and a solvent control.</li> </ol>
Toxicant Mixing:  1. Mixing chamber recommended but not required.	1. Mixing chambers were used
<ol> <li>Test solution completely mixed before introduction into the test system (aeration should not be used for mixing).</li> <li>Flow splitting accuracy must be within 10% and periodically checked.</li> </ol>	<ol> <li>Solution in mixing chamber was continuously stirred using a magnetic stirrer.</li> <li>Measured flows were within 4% of nominal.</li> </ol>

Guideline Criteria	Reported Information		
Exposure System/Test Vessels: Exposure tanks should be all glass or glass with a plastic or stainless steel frame (30.5 x 30.5 x 91.4 cm or 30.5 x 30.5 x 61 cm for fathead, and 45 x 90 x 26 cm for sheephead).	Each duplicate glass exposure aquarium was 60 x 30 x 30 cm, with two larval growth chambers (30 x 13 x 25 cm) placed in each aquarium.		
Larval chambers should have glass bottoms and drains that allow water to be drawn down to 3 cm.	Larval chamber material and drain height were not reported.		
Test water depth in adult tanks and larval chambers should be a minimum of 15 cm.	Test solution volume maintained at 27 L, equivalent to a solution depth of 15 cm.		
Embryo and Fry Chambers: 120 ml glass jars with bottoms replaced with 40 mesh stainless steel or nylon screen. Chambers can be oscillated vertically using rocker arm apparatus (2 rpm motor) or placed in separate chambers with self- starting siphons.	Embryo incubation cups were 5 cm diameter glass jars (height not reported) with 40-mesh Nitex® nylon screen bottoms. Two cups were placed within each incubation chamber (7.5 x 16 x 7.5 cm). During each diluter cycle, test solution flowed into and passed through the incubation chamber by means of a self-starting cap-siphon.		

Guideline Criteria	Reported Information		
Flow Rate: Flow rates to larval cups should provide 90% replacement in 8-12 hours, and maintain DO at above 75% of saturation. The toxicant level cannot drop below 20% with fish in the tank.	Flow rate to exposure tanks during the growth phase (Days 0-161) was 9.3 vol/24 hours (90% replacement in 6 hours). Flow rate to spawning tanks (Days 161-278) was 6.5 vol/24 hours (90% replacement in 8 hours).  DO levels ranged from 4.8 - 9.4 mg/L (58 - 114% of saturation); instances of DO levels <75% were corrected within 24 hours. Mean DO levels levels ranged from 84 to 91% of saturation.		
	Toxicant levels remained constant throughout the duration of the study; mean recoveries were 53 - 70% of nominal.		
Aeration: Dilution water should be aerated to insure DO concentration at or near 100% saturation. Test tanks and embryo chambers should not be aerated.	Between days 19 and 161, an oxygen saturating column was used to oxygenate the dilution water prior to entering the diluter cells. Between Days 161 and 278, gentle aeration was added to all aquaria.		

# C. Chemical System:

Guideline Criteria	Reported Information			
Concentrations: Minimum of 5 concentrations and a control, all replicated; plus solvent control if appropriate.  Toxicant conc. must be measured in one tank at each toxicant level every week.	A dilution water control, solvent control and 5 treatment concentrations: 1.3, 2.5, 5.0, 10, and 20 µg/L.  Test solutions were collected at least once per week from each replicate aquarium of the controls and treatments for analysis by GC until the initiation of the spawning period (Day 161). From Day 161 until test termination, samples were taken at least weekly from alternating replicates of each control and treatment for analysis by GC.			
Other Variables:  1. DO must be measured at each conc. at least once a week.  2. Test water temp. must be recorded continuously.  3. Freshwater: A control and one conc. must be analyzed weekly for pH, alkalinity, hardness, and conductance.  Natural seawater: must maintain a constant salinity and not fluctuate more than 6% weekly; monthly pH range <0.8 pH units.	<ol> <li>DO was measured daily in alternating replicate aquaria at each treatment level and control.</li> <li>Temperature was measured daily in alternating replicate aquaria at each treatment level and control. Temperature was also measured continuously in one exposure aquaria and one spawning aquaria using several minimum-maximum thermometers.</li> <li>Total hardness, alkalinity, and conductivity were measured weekly in one control aquarium and one treatment level aquarium on a rotating basis.</li> </ol>			

Guideline Criteria	Reported Information		
Solvents: Should not exceed 0.1 ml/L in a flow-through system. Acceptable solvents are: dimethylformamide, triethylene glycol, methanol, acetone, ethanol.	Solvent conc.: 11.5 µL/L Solvent: acetone		

# 12. REPORTED RESULTS:

Guideline Criteria	Reported Information
<ul> <li>Data Endpoints must include:</li> <li>survival of F<sub>0</sub> and F<sub>1</sub> embryos, time required to hatch, and hatching success;</li> <li>survival and total length of F<sub>0</sub> fish at 4 and 8 weeks after hatching;</li> <li>weights and lengths of F<sub>1</sub> fish at 8 weeks;</li> <li>incidence of pathological or histological effects; and</li> <li>observations of other effects or clinical signs.</li> </ul>	All biological parameters listed at left except:  The weights and lengths of F <sub>1</sub> fish were measured at 30 days post-hatch.

F<sub>0</sub> Results:

Nominal Conc. (µg/L)	Mean Measured Conc. (μg/L) (SD)	% Hatch	30-Day Post-Hatch % Survival	ost-Hatch Post-Hatch		
Control	<mdl*< td=""><td>88</td><td>89</td><td>89</td><td>88</td></mdl*<>	88	89	89	88	
Solvent Control	/ <b><m< b="">DL</m<></b>	89	85	85	90	
1.3 .6	0.69 (0.22)	85	94	94	100	
2.5 /. <sup>L</sup>	1.4 (0.3)	80	89	89	100	
5.02.9	2.9 (0.7)	85	86	86	96	
10.064	6.4 (1.2)	83	81	81	90	
20.0 14	14 (2.0)	63**	32**	32**	62**	

<sup>\*</sup> Minimum Detection Limit = 0.0954  $\mu$ g/L, mean procedural recovery = 103%.

<sup>\*\*</sup> Statistically different from the solvent control.

Mean Measured	Mean Total Length (mm)			Wet Weight (g)			
Concentration (µg/L)	30-Day	61- Day	278- Day (Male)	278- Day (Fem.)	61-Day	278- Day (Male)	278- Day (Fem.)
Control	31	40	86	70	0.563	8.3	4.1
Solvent Control	30	41	87	66	0.613	8.5	3.3
0.69	31	40	84	67	0.569	7.8	3.2
1.4	31	40	87	66	0.584	8.3	3.2
2.9	31	41	85	66	0.664	7.6*	3.1
6.4	29	40	83	65	0.608	7.2*	3.1
14	28*	41	81	67	_**	6.9*	3.5

<sup>\*</sup> Statistically different from the solvent control.

 $<sup>^{**}</sup>$  No 61-day weight data were reported for the 14  $\mu g/L$  treatment due to reduced survival (no fish were terminated).

Mean Measured Concentration (µg/L)	Number of Spawns	Total Number of Eggs	Number of Eggs/ Spawn	Number of Spawns/ Female	Number Eggs/ Female
Control	38	3942	104	8	841
Solvent Control	51	3810	73	9	679
0.69	63	6333	98	10	1056
1.4	37	2850	80	6	475
2.9	39	3233	83	7	539
6.4	14	503	35	2*	84
14	31	2648	75	5*	442

700

# F<sub>1</sub> Results:

Trams Me	Yeromes from 1	% Hatch	30-Day Post- Hatch % Survival	30-Day Post-Hatch Length (mm)	30-Day Post-Hatch Wet Weight (g)
	3.4-14 syle	85	92	30	0.25
	Control	88	94	30	0.26
	0.69	89	89	30	0.26
	1.4	78*	76	30	0.26
	2.9	76*	95	30	0.25
	6.4	93**	92**	29	0.23*
	14	27*	80**	26*	0.17*

<sup>\*</sup> Statistically different from the solvent control.

<sup>\*</sup> Significantly different from the solvent control.

<sup>\*\*</sup> No embryos incubated or larval groups exposed in replicate B due to reduced spawning activity. Therefore the data for this parameter at this treatment level were not included in the statistical analysis due to the lack of replication.

Toxicity Observations: Physical and behavioral abnormalities noted in the  $F_0$  fish included scoliosis, cranial malformations, and protruding eyes. Five cases of scoliosis were observed, two in the dilution water control, one in the solvent control, and two in the 0.69  $\mu$ g/L treatment. Three cranial malformations were noted, two in the dilution water control and one in the 6.4  $\mu$ g/L treatment. Two cases of protruding eyes occurred, one in the 1.4  $\mu$ g/L treatment and one in the 6.4  $\mu$ g/L treatment. One case of scoliosis was observed within the  $F_1$  fish.

# **Statistical Results:**

Statistical Method: Williams' test for growth and reproduction; were used for continuous data (length and weight) and Fisher's Exact test was used for survival and hatching success. All treatment data were compared to those of the pooled control.

Biological Endpoint	NOEC (μg/L)	LOEC (µg/L)
F <sub>0</sub> hatching success	6.4	14
F <sub>0</sub> 30-day survival	6.4	14
F <sub>0</sub> 30-day length	2.9	6.4
F <sub>0</sub> 61-day survival	6.4	14
F <sub>0</sub> 61-day length	>6.4	14
F <sub>0</sub> 61-day weight	>6.4	14
F <sub>0</sub> 278-day survival	6.4	14
F <sub>0</sub> 278-day length (Males)	2.9	6.4
F <sub>0</sub> 278-day length (Females)	6.4	>6.4
F <sub>0</sub> 278-day weight (Males)	2.9	7.5 6.4
F <sub>0</sub> 278-day weight (Females)	6.4 //	>6.4
F <sub>o</sub> # of spawns/female	2.9	6.4
F <sub>0</sub> # of eggs/female	14	>14
${ t F_1}$ hatching success	0.69	1.4
F <sub>1</sub> 30 day survival	6.4	14
F <sub>1</sub> 30 day length	2.9	>2.9
F <sub>1</sub> 30 day weight	2.9	>2.9

**NOEC:** 0.69  $\mu$ g/L **LOEC:** 1.4  $\mu$ g/L **MATC:** 0.98  $\mu$ g/L

### 13. REVIEWER'S STATISTICAL RESULTS:

Statistical Method: Bonferroni's Test and Dunnett's Test were used for continuous data (length and weight) and Williams' Test was used for survival and hatching success. All comparisons were made against the solvent control.

Biological Endpoint	NOEC (µg/L)	LOEC (µg/L)
F <sub>0</sub> hatching success	6.4	14
F <sub>0</sub> 30-day survival	6.4	14
F <sub>0</sub> 30-day length	6.4	14
F <sub>0</sub> 61-day survival	6.4	14
F <sub>0</sub> 61-day length	14	>14
F <sub>0</sub> 278-day survival	6.4	14
$F_0$ 278-day length (Males)	2.9	6.4
$F_0$ 278-day length (Females)	14	>14
F <sub>0</sub> 278-day weight (Males)	1.4	2.9
$F_0$ 278-day weight (Females)	14	>14
F <sub>0</sub> # of spawns/female	2.9	6.4
$F_0$ # of eggs/female	14	>14
F <sub>1</sub> hatching success	0.69	1.4
$F_1$ 30 day survival	6.4	14
F <sub>1</sub> 30 day length	6.4	14
F <sub>1</sub> 30 day weight	2.9	6.4

Most sensitive endpoint(s):  $F_1$  hatching success

**NOEC:** 0.69 ppb **LOEC:** 1.4 ppb **MATC:** 0.98 ppb

<u>Comments</u>: No data were reported for the 61-day weight of  $F_0$  fish in the 14  $\mu g/L$  treatment. Statistical analyses of 30-day  $F_1$  larval survival using Williams' test was not possible due to the limited degrees of freedom. The author's results were used for the NOEC and LOEC for this endpoint.

14. REVIEWER'S COMMENTS: This study is scientifically sound and fulfills the guideline requirements for a fish full life-cycle toxicity test using fathead minnows. The NOEC and LOEC for fathead minnows exposed to Technical Fluazinam were 0.69 and 1.4 ppb, respectively. The geometric mean MATC was 0.98 ppb. This study is classified as Core.

Technical Fluazinam - Fathead Life Cycle - F0 % Hatch File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS	TEST	(Isotonic	regression	model)	TABLE 1	OF	2

GROUP	IDENTIFICATION	N 	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Solvent	2	0.890	1.238	1.238
2	0.69	2	0.845.	1 <b>.1</b> 67	1.167
3	1.4	2	0.795	1.102	1.139
4	2.9	2	0.850	1.174	1.139
5	6.4	2	0.825	1.140	1.139
6	14	2	0.630	0.917	0.917

Technical Fluazinam - Fathead Life Cycle - F0 % Hatch File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TEST (Isotonic regression model) TABLE 2 0	WILLIAMS TEST	(Isotonic	regression	model)	TABLE 2	OF	2
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IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Solvent	1.238				
0.69	1.167	1.392		1.94	k = 1, v = 6
1.4	1.139	1.937		2.06	k=2, $v=6$
2.9	1.139	1.937		2.10	k = 3, v = 6
6.4	1.139	1.937		2.12	k = 4, $v = 6$
14	0.917	6.248	*	2.13	k = 5, v = 6

s = 0.051

Note: df used for table values are approximate when v > 20.

Technical Fluazinam - Fathead Life Cycle - F0 30-day % Survival File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Solvent	2	0.845	1.166	1.246
2	0.69	2	0.940	1.327	1.246
3	1.4	2	0.890	1.241	1.241
4	2.9	2	0.860	1.191	1.191
5	6.4	2	0.810	1.120	1.120
6	14	2	0.320	0.601	0.601

Technical Fluazinam - Fathead Life Cycle - F0 30-day % Survival File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TES	T (Isotonic	regression	model)	TABLE 2	OF 2	
	TSOTONTOED	CALC	STG	ጥአ ይፒ ፑ	DECDEES	0

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Solvent	1.246				
0.69	1.246	1.176		1.94	k = 1, v = 6
1.4	1.241	1.100		2.06	k = 2, v = 6
2.9	1.191	0.361		2.10	k = 3, v = 6
6.4	1.120	0.678		2.12	k = 4, v = 6
14	0.601	8.286	*	2.13	k = 5, v = 6

s = 0.068

Note: df used for table values are approximate when v > 20.

Fluazinam - F0 % Survival at Termination

File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

	WILLIAMS TEST (Isoto	nıc	regression mode.	1) TABLE 1 O	F 2
GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Solvent	2	0.900	1.251	1.397
2	0.69	2	1.000	1.471	1.397
3	1.4	2	1.000	1.471	1.397
4	2.9	2	0.960	1.377	1.377
5	6.4	2	0.900	1.251	1.251
6	14	2	0.620	0.908	0.908

Fluazinam - F0 % Survival at Termination

File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TEST	(Isotonic	regression	model)	TABLE 2 (	OF 2
IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Solvent 0.69 1.4 2.9 6.4 14	1.397 1.397 1.397 1.377 1.251 0.908	2.090 2.090 1.806 0.000 4.885	*\{ a *\}	1.94 2.06 2.10 2.12 2.13	k= 1, v= 6 k= 2, v= 6 k= 3, v= 6 k= 4, v= 6 k= 5, v= 6

s = 0.070

Note: df used for table values are approximate when v > 20.

a = Significantly higher than The solvent control.

Fluazinam - # Spawns/Female

File: 44807329 Transform: NO TRANSFORM

	WILLIAMS TEST (Isoto	nic	regression model	) TABLE 1 OI	F 2
GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1 2	Solvent 0.69	2 2	9.500 10.500	9.500 10.500	10.000
3	1.4	2	6.500	6.500	6.500
4	2.9	2	6.500	6.500	6.500
5	6.4	2	2.500	2.500	3.750
6	14	2	5.000	5.000	3.750

Fluazinam - # Spawns/Female

File: 44807329 Transform: NO TRANSFORM

WILLIAMS TEST	(Isotonic	regression	model)	TABLE 2 O	F 2
IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Solvent 0.69 1.4 2.9 6.4 14	10.000 10.000 6.500 6.500 3.750	0.285 1.708 1.708 3.275 3.275	* *	1.94 2.06 2.10 2.12 2.13	k= 1, v= 6 k= 2, v= 6 k= 3, v= 6 k= 4, v= 6 k= 5, v= 6

s = 1.756

Note: df used for table values are approximate when v > 20.

Fluazinam - # Eggs/Female File: 44807329 Transform: NO TRANSFORM

	WILLIAMS TEST (Isotor	nic	regression mod	del) TABLE 1 O	· 2
GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Solvent	2	679.000	679.000	867.250
2	0.69	2	1055.500	1055.500	867.250
3	1.4	2	475.000	475.000	507.000
4	2.9	2	539.000	539.000	507.000
5	6.4	2	84.000	84.000	262.750
6	14	2	441.500	441.500	262.750

Fluazinam - # Eggs/Female File: 44807329 Transform: NO TRANSFORM

WILLIAMS TEST	(Isotonic	regression	model)	TABLE 2 O	F 2
IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Solvent 0.69 1.4 2.9 6.4	867.250 867.250 507.000 507.000 262.750 262.750	0.637 0.582 0.582 1.408		1.94 2.06 2.10 2.12 2.13	k= 1, v= 6 k= 2, v= 6 k= 3, v= 6 k= 4, v= 6 k= 5, v= 6

s = 295.691

Note: df used for table values are approximate when  $v\,>\,20\,.$ 

Fluazinam - F1 % Hatch

File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Solvent	2	0.885	1.227	1.227
2	0.69	2	0.885	1.225	1.225
3	1.4	2	0.780	1.088	1.088
4	2.9	2	0.760	1.059	1.059
5	14	2	0.265	0.540	0.540

Fluazinam - F1 % Hatch

File: 44807329 Transform: ARC SINE(SQUARE ROOT(Y))

WILLIAMS TEST	(Isotonic	regression	model)	TABLE 2	OF	2

IDENTIFICATION	ISOTONIZED	CALC.	SIG	TABLE	DEGREES OF
	MEAN	WILLIAMS	P=.05	WILLIAMS	FREEDOM
Solvent 0.69 1.4 2.9	1.227 1.225 1.088 1.059 0.540	0.019 2.162 2.608 10.668	* * *	2.02 2.14 2.19 2.21	k= 1, v= 5 k= 2, v= 5 k= 3, v= 5 k= 4, v= 5

s = 0.064

Note: df used for table values are approximate when v > 20.

File:44807	73F0.s Tech	as Page 1 nical Fluazinam	n: Fathead Min 10:23	now FO Life Cyc Thursday, Sept	cle tember 30, 1999	File:448073F0.sas TMALEN 27 TFEMLEN 21
			TRT=1.3	•••••		TMALWT 27 TFEMWT 21
		Mean				
		30.5280899 0.5690744 39.6046512 83.5107692 66.5300000 7.6583500 3.1969917				Variable N THDLEN 80 SDWT 35 SDLEN 35 TMALEN 26 TFEMLEN 18 TMALWT 26
			TRY=10			TFEMWT 18
Variable	N	Mean	Std Dev	Minimum	Maximum	Technic
THDLEN SDWT SDLEN TMALEN TFEMLEN TMALWT TFEMWT	74 31 31 22 23 22 23	29.2972973 0.6091258 40.2580645 82.7604545 65.6334783 7.1528545 3.0515565	2.5196745 0.1502169 3.1301517 5.6369398 5.8886166 1.4694040 0.6556998	20.0000000 0.2809000 33.000000 69.200000 57.770000 3.9670000 1.5652000	34.0000000 0.8830000 46.0000000 98.9200000 86.9000000 10.5090000 4.0551000	Class Levels
						REP 24
Variable	N	Mean	Std Dev	Minimum	Maximum	Nu
		31.0500000 0.5847000 40.0000000 86.6730435 66.2242308 8.3268522 3.1625269	3.4673888 0.1593163 3.1119464 3.1118679 3.2425585 1.0109323 0.5691127	19.000000 0.3078000 33.0000000 81.3600000 58.6300000 6.8346000 2.0823000	50.0000000 0.9096000 45.0000000 92.0900000 72.1200000 11.0547000 4.6132000	
Variable	N	Mean	Std Dev	Minimum	Maximum	
THDLEN SDWT SDLEN TMALEN TFEMLEN TMALWT TFEMWT	31 0 17 15 15 15	27.7741935		21.0000000		NOTE: Variables in presence or Technic
	Techn	nical Fluazinam	: Fathead Minn 10:23	ow FO Life Cyc Thursday, Sept	le ember 30, 1999	Dependent Variable
			TRT=5			Source
Variable	N	Mean	Std Dev	Minimum	Maximum	Model
THDLEN SDWT SDLEN	85 36 36	30.6352941 0.6631278 41.4166667	2.8066548 0.1613478 2.9507868	22.0000000 0.4200000 36.0000000	40.0000000 1.2577000 48.0000000	Error Corrected Total

File:4480 TMALEN TFEMLEN TMALWT TFEMWT	073F0.sas 27 21 27 27 21	Page 2 84.4418519 66.1719048 7.6088630 3.1120238	4.6204004 4.4393531 1.2123531 0.7397190	74.7100000 59.6300000 5.3084000 2.1722000	96.7600000 78.4800000 10.3786000 5.3055000
			- TRT≃Solvent		
	) N				
IMALWI	35 35 26 18 26	30.3250000 0.6128229 40.6857143 86.7865385 66.4877778 8.4166577 3.3026111	0.1378160 2.5641353 4.9362706 2.7608046 1.1019555	34.0000000 75.2500000 61.4000000 6.6583000	0.9974000 46.0000000 96.2000000 71.2400000 10.4703000
	Techni	cal Fluazina		now FO Life Cyc	
			inear Models P Level Informa	rocedure	
Class	Levels				
TRT	6	1.3 10 2.5	20 5 Solvent	(Nominal)	
REP	24	1Aa 1Ab 1B 4Aa 4Ab 4B	la 1Bb 2Aa 2Ab la 4Bb 5Aa 5Ab	2Ba 2Bb 3Aa 3Ab 5Ba 5Bb 6Aa 6Ab	3Ba 3Bb 6Ba 6Bb
	N	umber of obs	ervations in d	ata set = 463	
		Group Ob	s Dependent	Variables	
		1 43	9 THDLEN		
		2 18	34 SDWT		
		3 20	1 SDLEN		
		4 13	9 TMALEN		
		5 12	7 TFEMLEN TF	EMWT	
		6 13	9 TMALWT		
			are consisten missing values	t with respect	to the
	Techni	cal Fluazina	m: Fathead Min 10:23	now FO Life Cyc Thursday, Sept	le ember 30, 1999
		General L	inear Models P	rocedure	
Dependent	Variable	e: THDLEN	Sum of	Mean	
Source		DF	Squares		alue Pr > F
Model		23	539.78874	23.46908	2.83 0.0001
Error		415	3437.76023	8.28376	
Corrected	Total	438	3977.54897		

File:448073F0.sas	Page 3 R-Square	c.v.	Root MSE	THDL	EN Mean
	0.135709	9.528724	2.8782		30.205
Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	5 18	327.44627 212.34248	65.48925 11.79680	7.91 1.42	0.0001 0.1156
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 18	0.00000 212.34248	11.79680	1.42	0.1156

#### General Linear Models Procedure Least Squares Means

TRT	THDLEN LSMEAN
1.3 10 2.5 20 5 Solvent	Non-est Non-est Non-est Non-est Non-est

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: THDLEN

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 415 MSE= 8.28376 Critical Value of T= 2.95229

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

Co	TRT omparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneou Upper Confidence Limit	
2.5	- 5	-0.9089	0.4147	1.7383	***
2.5	- 1.3	-0.7872	0.5219	1.8310	
2.5	- Solvent	-0.6185	0.7250	2.0685	
2.5	- 10	0.3822	1.7527	3.1232	
2.5	- 20	1.4781	3.2758	5.0735	
5 5 5 5	- 2.5 - 1.3 - Solvent - 10 - 20	-1.7;83 -1.1815 -1.0133 -0.0130 1.0783	-0.4147 0.1072 0.3103 1.3380 2.8611	0.9089 1.3959 1.6339 2.6890 4.6439	***
1.3	- 2.5	-1.8310	-0.5219	0.7872	
1.3	- 5	-1.3959	-0.1072	1.1815	
1.3	- Solvent	-1.1060	0.2031	1.5122	

ile:448073F0.sas Page 4 1.3 - 10 1.3 - 20	-0.1060 0.9818	1.2308 2.7539	2.5676 4.5260	***
Solvent - 2.5 Solvent - 5 Solvent - 1.3 Solvent - 10 Solvent - 20	-2.0685 -1.6339 -1.5122 -0.3428 0.7531	-0.7250 -0.3103 -0.2031 1.0277 2.5508	0.6185 1.0133 1.1060 2.3982 4.3485	***
10 - 2.5 10 - 5 10 - 1.3 10 - Solvent 10 - 20	-3.1232 -2.6890 -2.5676 -2.3982 -0.2948	-1.7527 -1.3380 -1.2308 -1.0277 1.5231	-0.3822 0.0130 0.1060 0.3428 3.3410	***
20 - 2.5 20 - 5 20 - 1.3 20 - Solvent 20 - 10	-5.0735 -4.6439 -4.5260 -4.3485 -3.3410	-3.2758 -2.8611 -2.7539 -2.5508 -1.5231	-1.4781 -1.0783 -0.9818 -0.7531 0.2948	*** *** ***

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Dunnett's One-tailed T tests for variable: THDLEN

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 415 MSE= 8.28376 Critical Value of Dunnett's T= 2.254

Comparisons significant at the 0.05 level are indicated by \*\*\*\*\*.

C	TRT omparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit	3
2.5	- Solvent	-0.3006	0.7250	1.7506	***
5	- Solvent	-0.7002	0.3103	1.3207	
1.3	- Solvent	-0.7963	0.2031	1.2025	
10	- Solvent	-2.0739	-1.0277	0.0185	
20	- Solvent	-3.9232	-2.5508	-1.1785	

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

### General Linear Models Procedure

Variable:	SDWT				
	DF	Sum of Squares	Mean Square	F Value	Pr > F
	9	0.2200770	0.0244530	1.07	0.3871
	174	3.9756684	0.0228487		
Total	183	4.1957454			
R	-Square	c.v.	Root MSE		SDWT Mean
0	.052452	24.94939	0.1512		0.6059
	Total R	9 174	DF Sum of Squares 9 0.2200770 174 3.9756684 Total 183 4.1957454 R-Square C.V.	DF Squares Square Square 9 0.2200770 0.0244530 174 3.9756684 0.0228487 Total 183 4.1957454 R-Square C.V. Root MSE	Sum of Squares Square F Value 9 0.2200770 0.0244530 1.07 174 3.9756684 0.0228487  Total 183 4.1957454 R-Square C.V. Root MSE

File:448073F0.sas Source	Page 5 DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	4 5	0.1957418 0.0243352	0.0489354 0.0048670	2.14 0.21	0.0777 0.9566
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 5	0.0000000 0.0243352	0:0048670	0.21	0.9566

#### General Linear Models Procedure Least Squares Means

TRT	SDWT LSMEAN
1.3 10 2.5 5 Solvent	Non-est Non-est Non-est Non-est

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: SDWT

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 174 MSE= 0.022849 Critical Value of T= 2.84327

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

TRT Compar		Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
5 - 5 -	Solvent 10 2.5 1.3	-0.05172 -0.05130 -0.02091 -0.00304	0.05030 0.05400 0.07843 0.09405	0.15233 0.15931 0.17776 0.19114
Solvent - Solvent - Solvent - Solvent -	10 2.5	-0.15233 -0.10230 -0.07195 -0.05409	-0.05030 0.00370 0.02812 0.04375	0.05172 0.10970 0.12819 0.14159
10 -	5 Solvent 2.5 1.3	-0.15931 -0.10970 -0.07899 -0.06121	-0.05400 -0.00370 0.02443 0.04005	0.05130 0.10230 0.12784 0.14131
2.5 -	5 Solvent 10 1.3	-0.17776 -0.12819 -0.12784 -0.07941	-0.07843 -0.02812 -0.02443 0.01563	0.02091 0.07195 0.07899 0.11066
1.3 - !	5	-0.19114	-0.09405	0.00304

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1.3 - Solvent	-0.14159	-0.04375	0.05409
1.3 - 10	-0.14131	-0.04005	0.06121
1.3 - 2.5	-0.11066	-0.01563	0.07941

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Dunnett's One-tailed T tests for variable: SDWT

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 174 MSE= 0.022849 Critical Value of Dunnett's T= 2.172

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

TRT Comparison		Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit	
5	- Solvent	-0.02763	0.05030	0.12824	
10	- Solvent	-0.08467	-0.00370	0.07728	
2.5	- Solvent	-0.10457	-0.02812	0.04832	
1.3	- Solvent	-0.11849	-0.04375	0.03099	

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

#### General Linear Models Procedure

Dependent Variab	le: SDLEN	Com. a.f.	M		
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	11	123.11544	11.19231	1.23	0.2683
Error	189	1717.24277	9.08594		
Corrected Total	200	1840.35821			
	R-Square	C.V.	Root MSE	St	DLEN Mean
	0.066898	7.460561	3.0143		40.403
Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	5 6	78.086092 45.029350	15.617218 7.504892	1.72 0.83	0.1321 0.5511
Source	DF	Type III SS	Mean Square	F Value	Pr > f
TRT REP	0 6	0.000000 45.029350	7.504892	0.83	0.5511

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

> General Linear Models Procedure Least Squares Means

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	TRT	SDLEN
		LSMEAN
	1.3	Non-est
	10	Non-est
	2.5	Non-est
	20	Non-est
	5	Non-est
	Solvent	Non-est

General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: SDLEN

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 189 MSE= 9.085941 Critical Value of T= 2.97297

Comparisons significant at the 0.05 level are indicated by \*\*\*\*.

				•
	TRT parison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
5 5 5 5 5	- 20 - Solvent - 10 - 2.5 - 1.3	-2.1029 -1.3963 -1.0371 -0.6545 -0.2124	0.5343 0.7310 1.1586 1.4167 1.8120	3.1715 2.8582 3.3543 3.4879 3.8365
20 20 20 20 20 20	- 5 - Solvent - 10 - 2.5 - 1.3	-3.1715 -2.4526 -2.0802 -1.7221 -1.2897	-0.5343 0.1966 0.6243 0.8824 1.2777	2.1029 2.8459 3.3288 3.4868 3.8451
Solvent Solvent Solvent Solvent	- 20 - 10 - 2.5	-2.8582 -2.8459 -1.7826 -1.4008 -0.9591	-0.7310 -0.1966 0.4276 0.6857 1.0811	1.3963 2.4526 2.6379 2.7722 3.1212
10 10 10 10 10	- 5 - 20 - Solvent - 2.5 - 1.3	-3.3543 -3.3288 -2.6379 -1.8982 -1.4580	-1.1586 -0.6243 -0.4276 0.2581 0.6534	1.0371 2.0802 1.7826 2.4144 2.7648
2.5 2.5 2.5 2.5 2.5	- 5 - 20 - Solvent - 10 - 1.3	-3.4879 -3.4868 -2.7722 -2.4144 -1.5863	-1.4167 -0.8824 -0.6857 -0.2581 0.3953	0.6545 1.7221 1.4008 1.8982 2.3770
1.3 1.3 1.3 1.3	- 5 - 20 - Solvent - 10 - 2.5	-3.8365 -3.8451 -3.1212 -2.7648 -2.3770	-1.8120 -1.2777 -1.0811 -0.6534 -0.3953	0.2124 1.2897 0.9591 1.4580 1.5863

Technical Fluazinam: Fathead Minnow FO Life Cycle

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General Linear Models Procedure

Dunnett's One-tailed T tests for variable: SDLEN

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 189 MSE= 9.085941 Critical Value of Dunnett's T= 2.257

Comparisons significant at the 0.05 level are indicated by \*\*\*\*\*.

C	TRT omparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
5	- Solvent	-0.8843	0.7310	2.3462
20	- Solvent	-1.8149	0.1966	2.2082
10	- Solvent	-2.1059	-0.4276	1.2506
2.5	- Solvent	-2.2700	-0.6857	0.8986
1.3	- Solvent	-2.6301	-1.0811	0.4680

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

#### General Linear Models Procedure

I					
Dependent	Variable: TMALEN	c 4	Mann		
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	12	617.50263	51.45855	2.14	0.0184
Error	126	3025.04686	24.00831		
Corrected	Total 138	3642.54949			
	R-Square	c.v.	Root MSE	TMA	LEN Mean
	0.169525	5.799313	4.8998		84.490
Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	5 7	473.38124 144.12140	94.67625 20.58877	3.94 0.86	0.0023 0.5421
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 7	0.00000 144.12140	20.58877	0.86	0.5421

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

> General Linear Models Procedure Least Squares Means

IRI	I MALEN LSMEAN	
1.3	Non-est	
10	Non-est	

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2.5 Non-est
20 Non-est
5 Non-est
Solvent Non-est

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: IMALEN

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 126 MSE= 24.00831 Critical Value of T= 2.99220

Comparisons significant at the 0.05 level are indicated by \*\*\*\*.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit	3
Solvent - 2.5	-4.083	0.113	4.310	***
Solvent - 5	-1.684	2.345	6.373	
Solvent - 1.3	-0.791	3.276	7.342	
Solvent - 10	-0.221	4.026	8.273	
Solvent - 20	0.552	5.306	10.060	
2.5 - Solvent	-4.310	-0.113	4.083	***
2.5 - 5	-1.929	2.231	6.391	
2.5 - 1.3	-1.035	3.162	7.359	
2.5 - 10	-0.460	3.913	8.285	
2.5 - 20	0.327	5.192	10.058	
5 - Solvent	-6.373	-2.345	1.684	
5 - 2.5	-6.391	-2.231	1.929	
5 - 1.3	-3.097	0.931	4.960	
5 - 10	-2.530	1.681	5.892	
5 - 20	-1.760	2.961	7.683	
1.3 - Solvent	-7.342	-3.276	0.791	
1.3 - 2.5	-7.359	-3.162	1.035	
1.3 - 5	-4.960	-0.931	3.097	
1.3 - 10	-3.497	0.750	4.997	
1.3 - 20	-2.724	2.030	6.784	
10 - Solvent	-8.273	-4.026	0.221	
10 - 2.5	-8.285	-3.913	0.460	
10 - 5	-5.892	-1.681	2.530	
10 - 1.3	-4.997	-0.750	3.497	
10 - 20	-3.629	1.280	6.189	
20 - Solvent	-10.060	-5.306	-0.552	***
20 - 2.5	-10.058	-5.192	-0.327	
20 - 5	-7.683	-2.961	1.760	
20 - 1.3	-6.784	-2.030	2.724	
20 - 10	-6.189	-1.280	3.629	

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

General Linear Models Procedure

Dunnett's One-tailed T tests for variable: TMALEN

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NOTE: This tests controls the type 1 experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 126 MSE= 24.00831 Critical Value of Dunnett's T= 2.270

Comparisons significant at the 0.05 level are indicated by \*\*\*\*\*.

C	TRT omparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneou Upper Confidence Limit	
2.5 5 1.3 10	- Solvent - Solvent - Solvent - Solvent	-3.298 -5.401 -6.361 -7.248	-0.113 -2.345 -3.276 -4.026	3.071 0.712 -0.191 -0.804	***
20	- Solvent	-8.913	-5.306	-1.699	***

Technical Fluazinam: Fathead Minnow FO Life Cycle 10:23 Thursday, September 30, 1999

### General Linear Models Procedure

Dependent	Variable: TFEMLE				
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	13	307.33763	23.64136	1.36	0.1899
Error	113	1964.13478	17.38172		
Corrected	Total 126	2271.47241			
	R-Square	C.V.	Root MSE	TFE	MLEN Mean
	0.135303	6.290005	4.1691		66.282
Source	DF	Type I \$\$	Mean Square	F Value	Pr > F
TRT REP	5 8	17.73728 289.60035	3.54746 36.20004	0.20 2.08	0.9602 0.0432
Source	DF	Type III \$\$	Mean Square	F Value	Pr > F
TRT REP	0 8 Technical Fluaz	0.00000 289.60035 inam: Fathead M 10:	36.20004 Hinnow FO Life 23 Thursday,	Cvcle	0.0432 30, 1999

#### General Linear Models Procedure

Dependent Variab	le: TFEMWT	C	Maaa		
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	13	10.161883	0.781683	1.91	0.0360
Error	113	46.261610	0.409395		
Corrected Total	126	56.423492			
	R-Square	c.v.	Root MSE	TFE	MWT Mean
	0.180100	20.03375	0.6398		3.1938
i .					

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Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	5 8	1.7911097 8.3707730	0.3582219 1.0463466	0.88 2.56	0.5003 0.0134
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 8	0.0000000 8.3707730	1.0463466	2.56	0.0134

#### General Linear Models Procedure Least Squares Means

TRT	TFEMLEN	TFEMWT
	LSMEAN	LSMEAN
1.3	Non-est	Non-est
10	Non-est	Non-est
2.5	Non-est	Non-est
20	Non-est	Non-est
5	Non-est	Non-est
Solvent	Non-est	Non-est

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General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: TFEMLEN

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 113 MSE= 17.38172 Critical Value of T= 2.99888

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
20 - 1.3	-3.758	0.357	4.472
20 - Solvent	-3.972	0.399	4.770
20 - 2.5	-3.391	0.662	4.716
20 - 5	-3.512	0.715	4.941
20 - 10	-2.896	1.253	5.403
1.3 - 20	-4.472	-0.357	3.758
1.3 - Solvent	-3.856	0.042	3.941
1.3 - 2.5	-3.233	0.306	3.845
1.3 - 5	-3.378	0.358	4.094
1.3 - 10	-2.752	0.897	4.545
Solvent - 20	-4.770	-0.399	3.972
Solvent - 1.3	-3.941	-0.042	3.856
Solvent - 2.5	-3.570	0.264	4.097
Solvent - 5	-3.700	0.316	4.332
Solvent - 10	-3.080	0.854	4.789
2.5 - 20	-4.716	-0.662	3.391

ile:4480731 2.5 2.5 2.5 2.5 2.5	FO.sas Page 12 - 1.3 - Solvent - 5 - 10	-3.845 -4.097 -3.616 -2.988	-0.306 -0.264 0.052 0.591	3.233 3.570 3.721 4.170
5	- 20	-4.941	-0.715	3.512
5	- 1.3	-4.094	-0.358	3.378
5	- Solvent	-4.332	-0.316	3.700
5	- 2.5	-3.721	-0.052	3.616
5	- 10	-3.235	0.538	4.312
10	- 20	-5.403	-1.253	2.896
10	- 1.3	-4.545	-0.897	2.752
10	- Solvent	-4.789	-0.854	3.080
10	- 2.5	-4.170	-0.591	2.988
10	- 5	-4.312	-0.538	3.235

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General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: TFEMWT

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 113 MSE= 0.409395 Critical Value of T= 2.99888

Comparisons significant at the 0.05 level are indicated by \*\*\*\*\*.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
20 - Solvent	-0.5284	0.1424	0.8132
20 - 1.3	-0.3835	0.2480	0.8796
20 - 2.5	-0.3397	0.2825	0.9046
20 - 5	-0.3157	0.3330	0.9817
20 - 10	-0.2434	0.3934	1.0303
Solvent - 20	-0.8132	-0.1424	0.5284
Solvent - 1.3	-0.4927	0.1056	0.7039
Solvent - 2.5	-0.4483	0.1401	0.7284
Solvent - 5	-0.4257	0.1906	0.8069
Solvent - 10	-0.3528	0.2511	0.8549
1.3 - 20	-0.8796	-0.2480	0.3835
1.3 - Solvent	-0.7039	-0.1056	0.4927
1.3 - 2.5	-0.5087	0.0345	0.5776
1.3 - 5	-0.4884	0.0850	0.6583
1.3 - 10	-0.4145	0.1454	0.7053
2.5 - 20	-0.9046	-0.2825	0.3397
2.5 - Solvent	-0.7284	-0.1401	0.4483
2.5 - 1.3	-0.5776	-0.0345	0.5087
2.5 - 5	-0.5125	0.0505	0.6135
2.5 - 10	-0.4383	0.1110	0.6602
5 - 20	-0.9817	-0.3330	0.3157
5 - Solvent	-0.8069	-0.1906	0.4257
5 - 1.3	-0.6583	-0.0850	0.4884
5 - 2.5	-0.6135	-0.0505	0.5125
5 - 10	-0.5187	0.0605	0.6396

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10 - 20	-1.0303	-0.3934	0.2434
10 - Solvent	-0.8549	-0.2511	0.3528
10 - 1.3	-0.7053	-0.1454	0.4145
10 - 2.5	-0.6602	-0.1110	0.4383
10 - 5	-0.6396	-0.0605	0.5187

General Linear Models Procedure

Dunnett's One-tailed T tests for variable: TFEMLEN

NOTE: This tests controls the type 1 experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 113 MSE= 17.38172 Critical Value of Dunnett's T= 2.244

Comparisons significant at the 0.05 level are indicated by \*\*\*\*\*.

Co	TRT mparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
20	- Solvent	-2.872	0.399	3.670
1.3	- Solvent	-2.875	0.042	2.959
2.5	- Solvent	-3.132	-0.264	2.605
5	- Solvent	-3.321	-0.316	2.689
10	- Solvent	-3.799	-0.854	2.090

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General Linear Models Procedure

Dunnett's One-tailed T tests for variable: TFEMWT

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 113 MSE= 0.409395 Critical Value of Dunnett's T= 2.244

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

	TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
20	- Solvent	-0.3596	0.1424	0.6444
1.3	- Solvent	-0.5533	-0.1056	0.3421
2.5	- Solvent	-0.5803	-0.1401	0.3002
5	- Solvent	-0.6518	-0.1906	0.2706
10	- Solvent	-0.7029	-0.2511	0.2008

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General Linear Models Procedure

Dependent Variable: TMALWT
Sum of Mean
Source DF Squares Square F Value Pr >

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Model	12	58.812362	4.901030	3.44	0.0002
Error	126	179.444612	1.424164		
Corrected Total	138	238.256974			
li de la companya de	R-Square	c.v.	Root MSE	TMA	LWT Mean
	0.246844	15.44189	1.1934		7.7282
Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	5 7	41.370942 17.441420	8.274188 2.491631	5.81 1.75	0.0001 0.1033
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 7	0.000000 17.441420	2:491631	1.75	0.1033

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> General Linear Models Procedure Least Squares Means

TRT	TMALWT LSMEAN
1.3 10 2.5 20 5 Solvent	Non-est Non-est Non-est Non-est Non-est

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General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: TMALWT

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 126 MSE= 1.424164 Critical Value of T= 2.99220

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit	3
Solvent - 2.5	-0.9324	0.0898	1.1120	***
Solvent - 1.3	-0.2321	0.7583	1.7487	
Solvent - 5	-0.1734	0.8078	1.7890	
Solvent - 10	0.2294	1.2638	2.2982	
Solvent - 20	0.4620	1.6198	2.7776	
2.5 - Solvent	-1.1120	-0.0898	0.9324	
2.5 - 1.3	-0.3537	0.6685	1.6907	

					1999							
* *			* * *	* *	ır 30,			r for		**	s	* * * *
1.7312 2.2389 2.7151	0.2321 0.3537 1.0306 1.5399 2.0193	0.1734 0.2952 0.9317 1.4816 1.9619	-0.2294 -0.1091 0.5289 0.5696 1.5517	-0.4620 -0.3449 0.2963 0.3379 0.8397	Life Cycle day, September	a	le: TMALWT	entwise error a control.	ISE= 1.424164 270	indicated by	Simultaneous Upper Confidence Limit	0.6857 -0.0069 -0.0634 -0.4790 -0.7413
0.7180 1.1740 1.5300	-0.7583 -0.6685 0.0495 0.5055 0.8615	-0.8078 -0.7180 -0.0495 0.4560 0.8120	-1.2638 -1.1740 -0.5055 -0.4560 0.3560	-1.6198 -1.5300 -0.8615 -0.8120 -0.3560	Minnow FO 0:23 Thursc	s Procedur	for variabl	I experim s against	95 df= 126 Mg Dunnett's T= 2	level are in	Difference Between Means	-0.0898 -0.7583 -0.8078 -1.2638
-0.2952 0.1091 0.3449	-1.7487 -1.6907 -0.9317 -0.5289	-1.7890 -1.7312 -1.0306 -0.5696	-2.2982 -2.2389 -1.5399 -1.4816 -0.8397	-2.7776 -2.7151 -2.0193 -1.9619	nam: Fathead 1	Linear Model	iled I tests	rols the type all treatments	Confidence= 0.95 tical Value of Dur	at the 0.05	Simultaneous Lower Confidence Limit	-0.8653 -1.5097 -1.5522 -2.0486 -2.4982
73F0.sas Page 15 - 5 - 10 - 20	- Solvent - 2.5 - 5 - 10 - 20	- Solvent - 2.5 - 1.3 - 10 - 20	- Solvent - 2.5 - 1.3 - 5	- Solvent - 2.5 - 1.3 - 5	Technical Fluazinam:	General	Dunnett's One-tai	: This tests control comparisons of all	Alpha= 0.05 confi Critical	significant	TRT Comparison	- Solvent - Solvent - Solvent - Solvent - Solvent
File:44807 2.5 2.5 2.5 2.5	 ผู้ผู้ผู้ผู้ผู้	លកសលក	55555	22222				NOTE	¥	Comparisons	5	2.5 1.3 20

	N				
THDLEN THDWT		Mean	Std Dev	Minimum	Maximum
	155 155	30.4047742 0.2610077	2.3779756 0.0577960		35.1200000 0.4136000
	· <b>-</b>		TRT=II		
Variable	N	Mean	Std Dev	Minimum	Maximum
THDLEN THDWT	91 91	29.5672527 0.2572560	1.6680615 0.0501795	23.8000000 0.1165000	33.0900000 0.3672000
			TRT=III		
Variable		Mean	Std Dev	Minimum	Maximum
THDLEN THDWT	142 142	29.7021831 0.2507711	2.3166169 0.0620282	20.7300000 0.0648000	34.0900000 0.3814000
			TRT=IV		
Variable	N	Mean	Std Dev	Minimum	Maximum
THDLEN THDWT	47 46	29.4106383 0.2295370	2.0611071 0.0628397	25.1300000 0.1314000	35.5900000 0.4645000
•••••			TRT=Solvent -		
Variable	N	Mean	Std Dev	Minimum	Maximum
THDLEN THDWT	141 140	29.8889362 0.2583421	1.5869552 0.0492993	24.9700000 0.1347000	33.5000000 0.3876000
			TRT=V		
Variable	N	Mean	Std Dev	Minimum	Maximum
THDLEN THDWT	22 20	25.8768182 0.1662250	3.8359309 0.0762581	16.4300000 0.0390000	32.4400000 0.3383000
	Techn	ical Fluazinam:		ow F1 Life Cyc ednesday, Sept	
			near Models Pr evel Informat	ocedure	•
lass L	evels	Values			
RT	6	1 11 111 1V	Solvent V		
REP	24	1Aa 2Aa 2Ab 4Bc 5Aa 5Ab	3AC 3Aa 3Ab 3 5Ac 5Ba 58b 5	Ba 3Bb 4Aa 4Ab Bc 6Aa 6Ab 6Ac	4Ba 4Bb 6Ba 6Bb

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Number of observations in data set = 625

Group Obs Dependent Variables

1 598 THDLEN

2 594 THDWT

NOTE: Variables in each group are consistent with respect to the presence or absence of missing values.

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### General Linear Models Procedure

Dependent Variab	le: THDLEN	C f	Mann		
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	23	731.05296	31.78491	7.56	0.0001
Error	574	2411.86815	4.20186		
Corrected Total	597	3142.92112			
	R-Square	C.V.	Root MSE	THO	DLEN Mean
	0.232603	6.891588	2.0498		29.744
Source	DF	Type I SS	Mean Square	f Value	Pr > F
TRT REP	5 18	407.96471 323.08825	81.59294 17.94935	19.42 4.27	0.0001 0.0001
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 18	0.00000 323.08825	17.94935	4.27	0:0001

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#### General Linear Models Procedure Least Squares Means

TRT	THDLEN LSMEAN
I II III IV Solvent V	Non-est Non-est Non-est Non-est Non-est

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General Linear Models Procedure

Bonferroni (Dunn) I tests for variable: THDLEN

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NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 574 MSE= 4.201861 Critical Value of T= 2.94754

Comparisons significant at the 0.05 level are indicated by '\*\*\*!.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneou Upper Confidence Limit	
I - Solvent I - III I - II I - IV I - V	-0.1873 0.0007 0.0396 -0.0120 3.1514	0.5158 0.7026 0.8375 0.9941 4.5280	1.2190 1.4044 1.6354 2.0002 5.9045	***
Solvent - I	-1.2190	-0.5158	0.1873	***
Solvent - III	-0.5316	0.1868	0.9051	
Solvent - II	-0.4908	0.3217	1.1341	
Solvent - IV	-0.5394	0.4783	1.4960	
Solvent - V	2.6271	4.0121	5.3971	
III - I	-1.4044	-0.7026	-0.0007	***
III - Solvent	-0.9051	-0.1868	0.5316	
III - II	-0.6764	0.1349	0.9463	
III - IV	-0.7252	0.2915	1.3083	
III - V	2.4410	3.8254	5.2097	
II - I	-1.6354	-0.8375	-0.0396	***
II - Solvent	-1.1341	-0.3217	0.4908	
II - III	-0.9463	-0.1349	0.6764	
II - IV	-0.9287	0.1566	1.2419	
II - V	2.2550	3.6904	5.1259	
IV - I	-2.0002	-0.9941	0.0120	***
IV - Solvent	-1.4960	-0.4783	0.5394	
IV - III	-1.3083	-0.2915	0.7252	
IV - II	-1.2419	-0.1566	0.9287	
IV - V	1.9730	3.5338	5.0946	
V - I V - Solvent V - III V - II	-5.9045 -5.3971 -5.2097 -5.1259 -5.0946	-4.5280 -4.0121 -3.8254 -3.6904 -3.5338	-3.1514 -2.6271 -2.4410 -2.2550 -1.9730	*** *** *** ***

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General Linear Models Procedure

Dunnett's One-tailed T tests for variable: THDLEN

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 574 MSE= 4.201861 Critical Value of Dunnett's T= 2.279

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

Simultaneous Simultaneous
Lower Difference Upper
TRT Confidence Between Confidence

File:448073F1.sas Page 4 Comparison		Limit	Means	Limit	
I III	- Solvent - Solvent	-0.0278 -0.7422	0.5158 -0.1868	1.0595 0.3687	
îî	- Solvent	-0.9499	-0.3217	0.3065	
I V	- Solvent	-1.2651 -5.0830	-0.4783 -4 0121	0.3085	***

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#### General Linear Models Procedure

Dependent Variabl	e: THDWT	Sum of	Mean		
Source	DF	Squares	Square	F Value	Pr > F
Model	23	0.5560780	0.0241773	8.92	0.0001
Error	570	1.5447247	0.0027100		
Corrected Total	593	2.1008027			
	R-Square	c.v.	Root MSE	T	HDWT Mean
	0.264698	20.68020	0.0521		0.2517
Source	DF	Type I SS	Mean Square	F Value	Pr > F
TRT REP	.5 18	0.1912507 0.3648273	0.0382501 0.0202682	14.11 7.48	0.0001 0.0001
Source	DF	Type III SS	Mean Square	F Value	Pr > F
TRT REP	0 18	0.0000000 0.3648273	0:0202682	7.48	0:0001

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> General Linear Models Procedure Least Squares Means

TRT	THDW LSMEAI
I	Non-es
ΙΙ	Non-es
III	Non-est
I۷	Non-es
Solvent	Non-es
٧	Non-es

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General Linear Models Procedure

Bonferroni (Dunn) T tests for variable: THDWT

NOTE: This test controls the type I experimentwise error rate but generally has a higher type II error rate than Tukey's for all pairwise comparisons.

Alpha= 0.05 Confidence= 0.95 df= 570 MSE= 0.00271

| File:448073F1.sas Page 5 | Critical Value of T= 2.94763

Comparisons significant at the 0.05 level are indicated by '\*\*\*'.

TRT Comparison	Simultaneous Lower Confidence Limit	Difference Between Means	Simultaneous Upper Confidence Limit
I - Solvent I - II I - III I - IV I - V	-0.015226 -0.016513 -0.007588 0.005707 0.058324	0.002666 0.003752 0.010237 0.031471 0.094783	0.020557 0.024016 0.028062 0.057235 *** 0.131241 ***
Solvent - I Solvent - II Solvent - III Solvent - IV Solvent - V	-0.020557 -0.019576 -0.010705 0.002727 0.055436	-0.002666 0.001086 0.007571 0.028805 0.092117	0.015226 0.021749 0.025847 0.054883 *** 0.128798 ***
II - I II - Solvent II - III II - IV II - V		-0.003752 -0.001086 0.006485 0.027719 0.091031	0.016513 0.019576 0.027090 0.055479 0.128926 ***
	-0.025847	-0.010237 -0.007571 -0.006485 0.021234 0.084546	0.007588 0.010705 0.014120 0.047267 0.121195 ***
IV - I IV - Solvent IV - II IV - III IV - V	-0.054883 -0.055479	-0.031471 -0.028805 -0.027719 -0.021234 0.063312	-0.005707 *** -0.002727 *** 0.000041 0.004798 0.104412 ***
V - I V - Solvent V - II V - III V - IV	-0.128798 -0.128926 -0.121195	-0.094783 -0.092117 -0.091031 -0.084546 -0.063312	-0.058324 *** -0.055436 *** -0.053136 *** -0.047897 *** -0.022212 ***

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General Linear Models Procedure

Dunnett's One-tailed T tests for variable: THDWT

NOTE: This tests controls the type I experimentwise error for comparisons of all treatments against a control.

Alpha= 0.05 Confidence= 0.95 df= 570 MSE= 0.00271 Critical Value of Dunnett's T= 2.279

Comparisons significant at the 0.05 level are indicated by \*\*\*\*.

TRT Comparison		Simultaneous Lower Confidence Limit	s Difference Between Means	Simultaneous Upper Confidence Limit	
I	- Solvent	-0.011170	0.002666	0.016501	
II	- Solvent	-0.017065	-0.001086	0.014893	
III	- Solvent	-0.021704	-0.007571	0.006562	

File:448073F1.sas Page 6 IV - Solvent -0.048972 -0.028805 -0.008638 \*\*\* V - Solvent -0.120484 -0.092117 -0.063751 \*\*\*